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### Virulence Dynamics and Diversity of *Puccinia striiformis* Populations in Egypt during 2017/18 and 2018/19 Growing Seasons

Ashmawy, M. A.\*; A. A. Shahin; Samar M. Esmail and Hend Abd El-Naby

Wheat Disease Research Department, Plant Pathology Research Institute (PPRI), Agricultural Research Center (ARC), Egypt

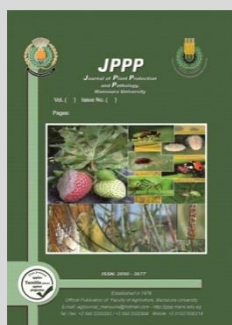


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#### ABSTRACT

Stripe rust, caused by *Puccinia striiformis* (Pst), is an important disease of wheat in Egypt. A total of nine and ten races were designated during two seasons. The most common race were 64E0, 66E0 and 70E20 (23.07%, 26.92% and 23.07) during 2017/18. While, race 450E254, 206E174, 192E192, 136E54 and 70E182 were the most common during 2018/19 at it was occurred at high frequency (22.50%, 17.5, 15.0, 10.0 and 10.0% frequencies, respectively). The three races; 64E0, 66E0 and 70E22 were the widely geographical distributed, as each was detected in all governorates during the first season, While, the 136E54, 192E192 and 206E174 during second season. No virulences were occurred to wheat stripe rust monogenic lines with Yr1, Yr5, Yr10 and Yr15, during the two years of the study. Different frequencies of virulence of Pst isolates were obtained against wheat cultivars used. Cultivars; Misr3, Giza171, Sakha94, Sakha95, and Gemmeiza12 showed the highest level of resistance against the tested Pst isolates. High similarity was found between stripe rust populations in the six locations. The phenotypic diversity within different populations was characterized using the three main indexes; Shannon, Gleason and Simpson. Shannon index proved to be more suitable to accurately measure the phenotypic diversity between the tested populations, as it was sensitive to sample size, number of isolates, number of races and standard deviation of race frequency than the others. Monitoring the dynamics and variation of virulence in stripe rust populations, that provides the basic information needed for an anticipatory breeding program for disease resistance in Egypt.

**Keywords:** Wheat, stripe rust, *Puccinia striiformis*, virulence dynamics, phenotypic diversity.



#### INTRODUCTION

Wheat stripe rust caused by *Puccinia striiformis* f. sp. *tritici* is the most destructive rust disease, causing a serious economic loss in grain yield of the susceptible wheat cultivars in Egypt, and worldwide (El-Daoudi *et al.*, 1996, Morgounov *et al.*, 2014 and Ashmawy and Rageb 2016). Since 2010, It has been considered to be a macrocyclic rust, after the identification of *Berberis* spp. as an alternative host and elucidation of the complete life history of this rust pathogen for the first time by Jin *et al.*, (2010).

Stripe rust was a dominant disease in Central Asian Countries in the late 1990s and early 2000s, accounting for yield losses of 20 and 40% in 1999 and 2000 in these countries (Morgounov *et al.*, 2014).

Historically, wheat stripe rust was considered a sporadic disease in Egypt, when during the last five decades, several stripe rust epidemics have been occurred at different intensities in 1967, 1983, 1986, 1995 and 1997, and more recently in 2018/19. These epidemics severely attacked the widely grown and high yielding wheat cultivars; Giza-144, Giza-150, Gemmeiza-1, Giza-163, Sakha-69, the long spikes; Sids cultivars and Sakha 93. Also, these severe epidemics were the main cause for eliminating and discarding the above wheat cultivars from agriculture and commercial production in Egypt (Abd El-Hak *et al.*, 1972, El-Daoudi *et al.*, 1996 and Abu El-Naga *et al.*, 2001).

Host-genetic resistance or growing wheat cultivars having a sustainable stripe rust resistance is still, the most effective, economically and environmentally safe control method, not only to minimize the annual crop loss, but also to avoid the sudden occurrence of severe epidemics in the future (Singh *et al.*, 2004). However, the amount of loss in grain yield depends, to a large extent, on the aggressiveness of the prevailing pathogen race(s), as well as the suitable environmental conditions favorable to rust incidence and development (Park *et al.*, 1988 and Hong and Singh 1996).

Successful control of stripe rust disease requires a full and thoroughly understanding of the pathogen races present in the local pathogen populations and the impact of the use of resistant cultivars on the frequencies of such races in its populations, and genetic structure of these populations all over the country. Thus, it is of major importance to conduct the annual survey of pathogen races throughout different wheat growing areas and during the successive growing seasons of wheat crop in Egypt. It is also, considered a very important step, required for evaluation and testing the genetic materials in the national breeding program for stripe rust resistance. Furthermore, analyzing virulence of stripe rust collections from different wheat growing areas in Egypt, and comparing diversity of races within these locations, represents an important prerequisite for understanding the genetic structure of these population in the country. Also, these informations

\* Corresponding author.

E-mail address: [dr\\_ashmawy2011@yahoo.com](mailto:dr_ashmawy2011@yahoo.com)  
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permit the decision markers to predict and avoid the sudden occurrence of the future disease epidemics and minimized and reduced the annual yield losses (Chen et al., 2009).

Therefore, the main objectives of this study were: 1) determine and identify frequency *P. striiformis* races in Egypt during 2017/18 and 2018/19 growing seasons; 2) study the geographical distribution of the identified Pst races in six Egyptian governorates; 3) estimate and measure the diversity and virulence dynamics between wheat stripe rust pathogen populations in different locations in Egypt.

## MATERIALS AND METHODS

### Field survey and sample's collection:

Wheat leaves naturally infected with stripe rust urediniospores were collected during the annual survey, in 2017/2018 and 2018/2019 growing seasons. These diseased samples were collected from both commercial fields and Egyptian wheat rust trap nurseries (EWRTN) grown in six governorates, i.e Kafrelsheikh, Beheira, Damietta, Dakahlia, Sharqia and Gharbia. Samples were air-dried by keeping each of them at room temperature (12 to 15°C) overnight in order to remove its moisture content. Samples were kept in paper pages (8 x 15 cm), and kept in the refrigerator at 2 to 5°C, until using in the isolation process.

### Isolation, purification and multiplication of urediniospores:

A susceptible wheat cultivar; Morocco was planted, as ten seeds per 10 cm diameter plastic pots in the greenhouse of Wheat Diseases Research Department, Plant Pathology Research Institute, ARC, Egypt. When the first leaf fully expanded in seven days old seedlings, leaves were rubbed gently between moist fingers with tap water. Then, infected samples were scraped using sterile spatula and sprayed gently again with water in order to form a film of free water, which is essential to initiate spore germination and establishment of infection. The inoculated seedlings were incubated in moist chambers for 24 h at 9 to 12°C and 100% RH. Then moved to greenhouse benches and kept for 14 days at approximately 12 ± 2°C. After pustule's rupture, three single pustules were isolated separately from each specimen for multiplication of spores on the highly susceptible variety; Morocco to, obtain enough urediniospores or generate sufficient inocula (spores), for race identification and nomenclature, as described by Johnson et al. (1972).

### Race designation and nomenclature:

Seventeen differential wheat genotypes; both world and European set of differentials, were used to identify and designate stripe rust races (Table, 1), following the method of Stakman et al. (1962).

The differential sets consisted of nine world differentials and eight European differential set of wheat lines possessing 17 stripe rust resistance genes, as presented in Table (1), and described by Johnson et al., (1972). Uridionosporos obtained from each pure isolate were used to inoculate the set of differentials, following the method of Stakman et al., (1962)

After seven-days, seedlings were inoculated by shaking and brushing rust spores with the previously

isolated single pustule isolates of *P. striiformis*. The inoculated seedlings were incubated in the humid chamber overnight (100% RH), as described above. The inoculated seedlings were transferred, onto the greenhouse benches. After 10-12 days, infection types (IT's) were recorded for all differential lines, using standard disease scoring scale of 0- 9 grades ( Mc Neal, 1971) Entries which showed low infection types (L), i.e. scores = 0, 0; , 1,2,3,4 and 5 were considered host resistant and avirulent isolates. While those showed high infection types (H), i.e. scores = 6,7,8 and 9, were recorded as the susceptible lines and virulent isolates.

**Table 1.\*Differential genotypes used to identify races of wheat stripe rust, incited by *Puccinia striiformis f. sp. tritici* in Egypt.**

Differential genotypes	Abbreviation	Decanery value	Resistance gene(s)	Type
GI. World differential set:				
Chinese 166	Ch	(2 <sup>0</sup> ) = 1	<i>Yr1</i>	winter
Lee	Lee	(2 <sup>1</sup> ) = 2	<i>Yr7</i>	spring
Heines Kolben	HK	(2 <sup>2</sup> ) = 4	<i>Yr2Yr6</i>	spring
Vilmorin 23	V23	(2 <sup>3</sup> ) = 8	<i>Yr3</i>	winter
Moro	Mo	(2 <sup>4</sup> ) = 16	<i>Yr10</i>	winter
Strubes Dickkopf	Std	(2 <sup>5</sup> ) = 32	<i>SD</i>	winter
Suwon 92 × Omar	Su	(2 <sup>6</sup> ) = 64	<i>SU</i>	winter
Clement	Cl	(2 <sup>7</sup> ) = 128	<i>Yr2Yr9</i>	winter
<i>Triticum spelta</i> Album	Sp	(2 <sup>8</sup> ) = 256	<i>Yr5</i>	spring
GII. European Differential set:				
Hybrid 46	H46	(2 <sup>0</sup> ) = 1	<i>Yr4</i>	winter
Reichersberg 42	R42	(2 <sup>1</sup> ) = 2	<i>Yr(7)</i>	winter
Heines Peko	Pe	(2 <sup>2</sup> ) = 4	<i>Yr2 Yr(6)</i>	spring
Nord Desprez	No.D	(2 <sup>3</sup> ) = 8	<i>Yr(3)</i>	winter
Compair	Com	(2 <sup>4</sup> ) = 16	<i>Yr8</i>	spring
Carstens V	CV	(2 <sup>5</sup> ) = 32	<i>YrCV</i>	winter
Spaldings Prolific	Spa	(2 <sup>6</sup> ) = 64	<i>YrSP</i>	winter
Heines VII	HVII	(2 <sup>7</sup> ) = 128	<i>Yr2</i>	winter

\* Johnson et al., (1972).

### Virulence frequency (%)

Frequency of virulence was estimated for each Pst race identified in the current study, as a percentage of virulent isolates to the total number of the tested isolates, according to the following equation:

$$\text{Virulence frequency}(\%) = \frac{\text{No. of virulent isolates}}{\text{Total number of isolates}} \times 100$$

### Measurements of race diversity:

The number and frequency of races within populations of *P. Striiformis f. Sp. tritici* obtained from six governorates in Egypt, during 2017/18 and 2018/19 growing seasons, were used to measure the diversity of races in each population of the different regions, under study. To assess and measure diversity of *P. striiformis* races within each population (region), three widely used indexes, i.e. Shannon index (H<sub>SH</sub>), Gleason index (H<sub>G</sub>) and Simpson index (H<sub>S</sub>). These three popular diversity indexes were calculated from data on races of *P. striiformis f. Sp. tritici* from different regions (locations) and years under study. To assess diversity of races within each population (region), three indexes, i.e. Shannon index (HSH), Gleason index (HG) and Simpson index (HS) (Groth and Roelfs, 1987) were estimated as follows.

**1- Shannon index( $H_{SH}$ ):** This index was used to determine the similarities of the frequencies of the different pathotypes in a set of isolates, by the following formula:

$$H_{SH} = -\sum (P_i \ln P_i)$$

**Where:**  $P_i$  is the frequency of the  $i$  th pathotype in the set of isolates.

**2- Gleason index( $H_G$ ):** Gleason index was used to detect the number of distinct pathotypes present, indicating the richness aspect of diversity, and calculated by the following formula:

$$H_G = (n-1)/\ln(N)$$

**Where:**  $n$  is the number of pathotypes, identified in the sample and  $N$  is the total number of isolates in the sample.

**3- Simpson index( $H_S$ ):** Simpson index was another popular diversity index for plant pathogens to determine the number of pathotypes and evenness of their distribution. It was calculated using the following formula:

$$H_S = 1 - \sum [n_i(n_i - 1)/N(N-1)]$$

**Where:**  $n_i$  is the number of isolates of the  $i$  th pathotype, while  $N$  is the sample size.

### Statistical analysis

Correlation and regression coefficient “SPSS Regression Modeling” was used to determine the relationship between each of the four components of diversity, i.e. sample size collected from each governorate(region), number of the obtained isolates, number of the identified *Pst* races, and standard deviation of race frequency, and each of the three diversity indexes, i.e. Shannon, Gleason and Simpson, over the two growing seasons of the study.

## RESULTS AND DISCUSSION

### Results

During the annual survey of wheat stripe rust in the two growing seasons 2017/18 and 2018/19, a total of 250 samples were collected from the six Egyptian governorates under study Kafrelsheikh, Gharbia, Dakahlia, Damietta, Behera and Sharqia. These samples produced 330 single isolates of the causal pathogen (Table 2).

In 2017/18 growing season, 100 samples were collected from the different locations, under study. These samples produced 130 single isolates. The highest number of the collected samples was obtained from Kafrelsheikh (27 samples), while the lowest number of these samples were obtained from Dakahlia (only 8 samples). The highest number of stripe rust isolates was obtained from the samples collected from Kafrelsheikh (41 *Pst* isolates), followed by Gharbia and Behera, which produced 26 and

24 pure isolates, respectively. While, the lowest number of these isolates was obtained from Dakahlia, Sharqia and Damietta, which produced 11, 13 and 15 *Pst* isolates, respectively (Table 2).

A total of 200 pure isolates of the causal pathogen were obtained from 150 samples, collected from the six Egyptian governorates in 2018/19 growing season, (Table 2). As it was found in the first season, the highest numbers of the collected samples were obtained from Kafrelsheikh, Beheira and Gharbia, i.e. 59, 32 and 22 samples, respectively, while the lowest numbers of these samples were obtained from Damietta, Dakahlia and Sharqia i.e. 11, 13, 13 samples, respectively. Moreover,

### Race occurrence and their frequencies (%) in pathogen populations:

Based on infection type (IT ) on the series of 17 different tail lines of *Pst*, both world and European ones (Table 2), 9 and 10 *Pst* races were identified during 2017/18 and 2018/19 growing seasons, respectively (Table 3). The nine virulent races of *Puccinia striiformis* that identified and designated from a total of 130 pure isolates in Egypt during 2017/18 growing season, were 0E0, 0E16, 4E130, 64E0, 66E0, 70E27, 70E32, 70E182 and 70E214. As indicated in Table 3, race 64E0 is the most common race, where, it showed the highest frequency (26.92%), followed by the four races; 66E0, (23.07%), 70E26 (15.38%), 70E182 (11.53%), and 4E130(7.69%), respectively. While, the other stripe rust races under study, i.e. 0E0, 0E16, 70E32 and 70E214, were rare as they showed relatively low frequencies, each represented by only 4 isolates in pathogen population with 3.84 % frequency for each (Table 3).

In the second growing season i.e. 2018/19, ten *P. striiformis* races were designated from a total of 200 pure isolates. Races; 450E254 occurred at a consistently high frequency in its population during this season; where, it was found at 22.5 % frequency, followed by the two races i.e. 206E174 and 192E192 as they showed 17.5% and 15.0% frequencies. Seven of the identified *Pst* races found at the relatively low frequencies in the pathogen population (ranged from 2.5% to 10.0% frequency) (Table 3).

Four *Pst* races, i.e. 0E0, 0E16, 4E130 and 70E182 were common and have been detected and found in all collections during the two growing seasons of the study, in all *Pst* collections but with different frequencies in these seasons. Meanwhile, some of stripe rust races were found in only one season, but they did not found in the other season. For example, the five races; 64E0, 66E0, 70E26, 70E32 and 70E214.

**Table 2. Number of stripe rust samples and isolates obtained from different wheat growing areas in Egypt during 2017/18 and 2018/19 growing seasons.**

No.	Locations	Gerowing season / Number of samples and isolates					
		2017/2018		2018/2019		Total No. of	
		No. Of samples	No. Of isolates	No. Of samples	No. Of isolates	samples	Isolates
1-	Kaferelsheikh						
	Biala	10	18	19	23	29	41
	Qualin	5	8	3	4	8	12
	Sidi Salem	4	5	19	23	23	28
	Motobe	4	5	14	18	18	23
	Baltiam	4	5	4	6	8	11
	Total	27	41	59	74	86	115
2-	Gharbia						
	Qoutour	3	4	3	5	6	9
	Bassion	2	3	4	5	6	8
	El-Mahalla	5	6	4	6	9	12
	Zefta	6	7	4	6	10	13
	Tanta	5	6	7	9	12	15
	Total	21	26	22	31	43	57
3-	Damietta						
	El- Zarqua	2	4	2	4	4	8
	Kafr- Saad	3	5	4	7	7	12
	Faraskor	5	6	5	8	10	14
	Total	10	15	11	19	21	34
4-	Dakahlia						
	Mansoura	3	4	5	6	8	10
	Belqas	2	3	4	5	6	8
	Talkha	3	4	4	5	7	9
	Total	8	11	13	16	21	27
5-	Beheira						
	Damnhor	6	7	6	8	12	15
	Etag- elbarod	3	3	6	8	9	11
	El-Mahmodiya	4	4	7	9	11	13
	Hosh Esa	5	5	8	10	13	15
	Kafer el-Douar	5	5	5	8	10	13
	Total	23	24	32	43	55	67
6-	Sharqia						
	Abo-Kber	6	7	6	8	13	15
	Kafer-El hamam	5	6	7	9	11	15
	Total	11	13	13	17	24	30
	Total	100	130	150	200	250	330

samples collected from Kafrelsheikh and Beheira produced the highest number of single isolates, i.e. 74 and 43, respectively (Table 2).

**Table 3. *Puccinia striiformis* race, number of isolates and their frequency (%) within its populations in Egypt during 2017/18 and 2018/19 growing seasons.**

N0.	Pst races	2017/2018		2018/2019	
		No. of isolates	Frequency* (%)	No. of isolates	Frequency (%)
1	0E0	5	3.84	5	2.5
2	0E16	5	3.84	15	7.5
3	4E130	10	7.69	5	2.5
4	46E24	-	-	10	5.00
5	64E0	35	26.92	-	-
6	64E8	-	-	15	7.5
7	66E0	30	23.07	-	-
8	70E20	20	15.38	-	-
9	70E32	5	3.84	-	-
10	70E182	15	11.53	20	10.00
11	70E214	5	3.84	-	-
12	136E54	-	-	20	10.00
13	192E192	-	-	30	15.00
14	206E174	-	-	35	17.5
15	450E254	-	-	45	22.5
	Total	130	-	200	-

• Frequency (%) : means frequency of each Pst race within its population.

were detected in only the first growing season, and not in the second season. while, the four Pst races i.e. 13E54, 192E192, 206E174 and 450E254, have been appeared and detected for the first time in the second growing season (Table 3).

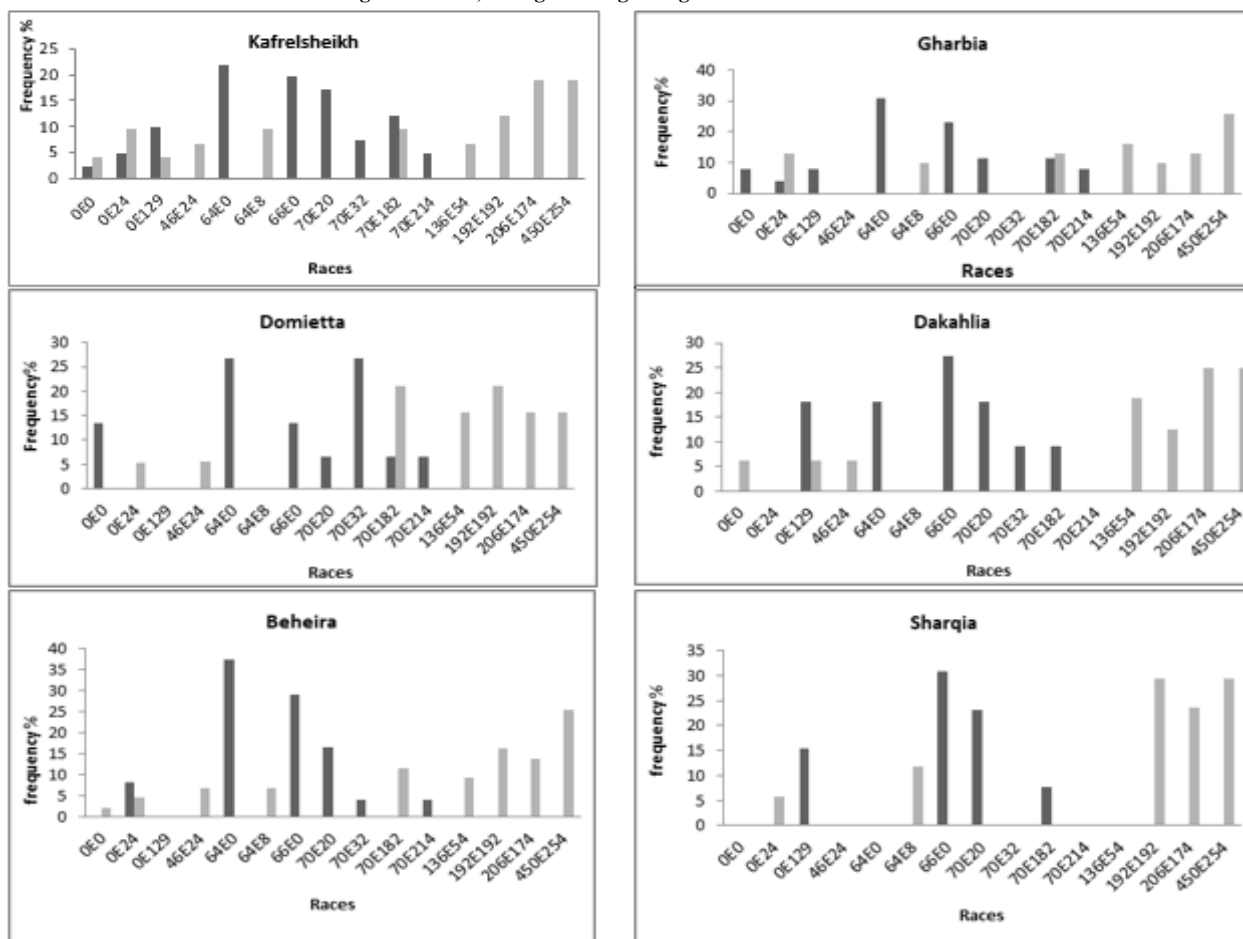
**Geographical distribution of stripe rust races:**

All Pst races detected in the two growing seasons under study, were also identified and found in Kaferelsheikh population. Thus, it is considered to be the largest population size, compared to the other pathogen populations in the tested governorates. As, it contains all the nine races of the casual pathogen identified in 2017/2018, i.e. 0E0, 0E16, 4E130, 64E0, 66E0, 70E26, 70E32, 70E182 and 70E214, (Table, 4 and Fig. 1). Also, the ten races; 0E0, 0E16, 4E130, 64E24, 64E8, 70E182, 136E54, 192E192, 206E174 and 450E254, which identified in Pst collections during 2018/2019, were also found among the collections from Kaferelsheikh, governorates (Table, 4 and Fig. 1). The obtained results during the first season revealed, on the other hand, that Pst race; 64E0 was the most frequent in this population (with 21.95% frequency) followed by the three Pst races; 66E0, 70E26 and 70E182, as they showed 19.51%, 17.07% and 12.19% frequencies in this season, respectively. While, in the second season the two races; 206E174 and 450E254 showed the relatively high frequency (18.91% for each), followed by 192E192 (12.16% frequency). The other Pst races were low in their frequencies (not exceeded up 9.45% ) within pathogen populations in Kaferelsheikh governorate, during the two growing seasons of the study (Table, 4 and Fig. 1).

**Table 4. Frequency(%) of *Puccinia striiformis* (Pst) races, collected from wheat growing areas of the six governorates in Egypt, during 2017/2018 and 2018/2019 growing seasons.**

Pst races	Frequency (%) of races collected from indicated area											
	Kaferelsheikh		Gharbia		Damietta		Dakahlia		Beheira		Sharqia	
	2017/18	2018/19	2017/18	2018/19	2017/18	2018/19	2017/18	2018/19	2017/18	2018/19	2017/18	2018/19
0E0	2.43	4.05	7.69	-	13.33	-	-	6.25	-	2.32	-	-
0E16	4.87	9.45	3.84	12.90	-	5.26	-	-	8.33	4.65	-	5.88
4E130	9.75	4.05	7.69	-	-	-	18.18	6.25	-	-	15.38	-
46E24	-	6.75	-	-	-	5.63	-	6.25	-	6.97	-	-
64E0	21.95	-	30.76	-	26.66	-	18.18	-	37.5	-	23.07	-
64E8	-	9.45	-	9.96	-	-	-	-	-	6.97	-	11.76
66E0	19.51	-	23.07	-	13.33	-	27.27	-	29.16	-	30.76	-
70E20	17.07	-	11.53	-	6.66	-	18.18	-	16.66	-	23.07	-
70E32	7.31	-	-	-	26.66	-	9.09	-	4.16	-	-	-
70E182	12.19	9.45	11.53	12.90	6.66	21.05	9.09	-	-	11.63	7.67	-
70E214	4.87	-	7.69	-	6.66	-	-	-	4.16	-	-	-
136E54	-	6.75	-	16.12	-	15.78	-	18.75	-	9.30	-	-
192E192	-	12.16	-	9.67	-	21.05	-	12.50	-	16.27	-	29.41
206E174	-	18.91	-	12.90	-	15.78	-	25.00	-	13.95	-	23.52
450E254	-	18.91	-	25.80	-	15.78	-	25.00	-	25.58	-	29.41
Total *	9	10	8	7	7	7	6	7	6	9	5	5

\*Total number of identified races in each governorates., during the two growing seasons.



**Fig. 1. Temporal and spatial variations in *Puccinia striiformis* races and geographical distribution within six local populations in Egypt during 2017/18 and 2018/19 grwing seasons.**

The obtained data in Table 4 and Fig. 1, showed, in contrast, that Sharqia Pst populations were the smallest population sizes, as they contains only 5 races in each season, under study. However, Pst race, 66E0 was the most frequent (with 30.76% frequency), followed by the two races; 64E0 and 70E26 (each with 23.07% frequency) during the first season. While, in the second season the

two, races; 192E192 and 450E254 showed the highest frequency (each with 29.41% frequency), followed by 206E174 , as it was found by 23.52% frequency (Table, 4and Fig. 1). In contrast, the other two races identified in Sharqia population were rare, as they showed onely 5.88% and 11.76% frequency in this population, respectively. Out of the tested pathogen populations, Pst populations of

the four governorates; Gharbia, Damietta, Dakahlia and Beheira showed in general moderate sizes of *Pst* populations, while they contained only 8, 7, 6 and 6 races in the first season, and 7, 7, 7 and 9 in the second season, respectively (Table 4 and Fig. 1).

**Virulence formula and Virulence frequency (%) of the tested races:**

**a. Virulence formula:**

Data presented in Table, (5) revealed that the most aggressive *Pst* races under study were the two races 450E254 and 206E174, as each was virulent to 11 genes

for stripe rust resistance from the total of the tested genes. However, race 206E174 found to be virulent to *Yr*'s; 7,6,3,*SU*,9,(7),(6),(3),*CV*, and 2. Also, race 450E254 proved to be virulent to *Yr*'s; 7,*SU*,9,5,(7),(6),(3),8,*CV*,*SP*, and 2. In contrast, race 0E0 didn't showed any virulence reaction against all of the tested yellow rust resistance genes (*Yr*'s). On the other hand, the rest of the identified *Pst* races showed moderate to low virulence frequencies (ranging from 5.88% to 47.08%) reaction against different *Yr* genes under study, as they have the ability to be virulent against at least 1, 2, 5, 6 and 8 *Yr* genes (Table 5).

**Table 5. Virulence formula, and virulence frequency (%) of *Puccinia striiformis* races (Pst) identified in Egypt during 2017/18 and 2018/19 growing seasons.**

No.	Race	Virulence formula (ineffective genes)	Growing season / Virulence formula & Virulence frequency (%) of races			
			2017/2018		2018/2019	
			No. of ineffective genes	Virulence frequency (%)	No. of ineffective genes	Virulence frequency (%)
1	0E0	-	0	0.0	0	0.0
2	0E16	8	2	11.76	2	11.76
3	4E130	6,(7),2	2	11.76	5	11.76
4	46E24	7,6,3, <i>SD</i> ,(3),8	-	-	6	35.29
5	64E0	<i>Su</i>	1	5.88	-	-
6	64E8	<i>SU</i> , (3)	-	-	2	11.76
7	66E0	7, <i>Su</i>	2	11.76	-	-
8	70E20	6,7, <i>Su</i> ,(7),(3),8	6	35.29	-	-
9	70E32	6,7, <i>Su</i> , <i>Cv</i>	4	23.53	-	-
10	70E182	6,7, <i>SU</i> ,(7),(6),8, <i>CV</i> ,2	8	47.08	8	47.08
11	70E214	6,7, <i>SU</i> ,(7),(6),8, <i>SP</i> ,2	8	47.08	-	-
12	136E54	7, 6, 9, (3),8, <i>CV</i>	-	-	6	35.29
13	192E192	<i>SU</i> ,9,(7),(6), (3),8, <i>CV</i> ,2	-	-	8	47.08
14	206E174	7,6,3, <i>SU</i> ,9,(7),(6),(3), <i>CV</i> ,2	-	-	11	64.70
15	450E254	7, <i>SU</i> ,9,5,(7),(6),(3),8, <i>CV</i> , <i>SP</i> ,2	-	-	11	64.70

<sup>a</sup>Virulence frequency (%) calculated by divided the number of ineffective genes for each race to the total number of resistance genes used (17 *Yr*'s) X 100.

**b- Virulence frequency (%):**

Frequency of virulence for each race was detected against the seventeen differential hosts for stripe rust resistance (17 *Yr*'s) in the two years under study. Occurrence of virulence frequency for each race was estimated as the percentage of the number of ineffective genes to the total number of resistance genes used (Table 5). Different virulence frequencies (%) of the tested races were occurred ranged from 0% for 0E0 pallyoty to 64.70% for each of the two *Pst*, 450E254 and 206E174 had the widest virulence spectrum, thus they were the most aggressive (*Pst*) races, where it proved to be virulent against with virulence the most differential lines a genotypes, In the same time, they showed the highest virulence frequencies (each with 64.70%), followed by the three races *i.e.* 70E182, 70E214 and 192E192 that possessing a rebtishly high virulence frequency reached to 47.08%. In contrast, race 0E0 displayed avirulent reaction or did not have the potentiality to be virulent against all the tested *Yr*'s, and it, therefore, showed 0.00% virulence frequency (Table 5).

Occurrence of virulence was also estimated as the percentage of virulent isolates to the total number of isolates for each wheat genotype under study, both yellow rust resistance genes (34 *Yr*'s) and 18 local wheat varieties (Tables 6 and 7).

**Virulence of *Pst* isolates against different wheat genotypes:**

**a- Virulence of the tested isolates to yellow rust resistance genes (*Yr*'s)**

Results obtained in the first season that presented in Table (6), showed ingeneral, different frequencies of virulence to

the tested stripe rust resistance lines (*Yr*'s). No virulences (0.0 % VF) of the *Pst* isolates were found against *Yr1*, *Yr5*, *Yr10*, *Yr15*, *Yr17*, *Yr27*, *Yr32* and *YrSp*, as were highly effectived and were completely resistance against all isolates under study (100.0% efficacy). On the other hand, the highest occurrence of for the *Pst* isolates virulence was found against *Yr2*, *Yr3*, *Yr4*, *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr18*, *YrA*, *YrND*, *YrSD* and *YrSU*, ranging from 53.84 to 100%. Whereas, the rest lines (*Yr*'s) under study showed moderate responses during this season. In the seacend season, the obtained results also revealed that the four yellow rust resistance genes *Yr*'s; *Yr1*, *Yr5*, *Yr10* and *Yr15* exhibited complete resistance against all the tested isolates, as they showed the lowest frequencies of virulence being was 0.0% all. In the same time, these *Yr*'s considered to be the highly effective (resistant) genes against all the prevalent *Pst* isolates in the collections of this season. On the other hand, according to the virulence frequency of *Yr*'s; *Yr17*, *Yr27*, *Yr32*, *YrSp*, *Yr3*, *Yr4*, *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr18*, *YrA*, *YrND*, *YrSD* and *YrSU* which ranged from 15.0% to 95.00%, these resistance genes (*Yr*'s) considered to be ineffective during 2018/19 growing season (Table 6)

**b- Virulence to the local wheat varieties:**

Data the presented in Table (7), revealed, in general, the 18 wheat varteties different levels of varietal resistance against the tested stripe rust isolates, obtained from the collections during the two growing seasons 2017/18 and 2018/19. The highest levels of varietal resistance to the tested *Pst* isolates was displayed in wheat cultivars *i.e.* Sakha94, Sakha95, Gemmeiza12, Misr2, Shandaweel 1, Giza171 and Misr3 (ranging from 92.40 to

96.16%) in an as order. On the other hand, the lowest levels of varietal resistance to the tested pathogen isolates were found in wheat cultivars; Sids12, Sakha69, Gemmeiza11, Sakha93 and Sids13 (ranging from 23.07% to 39.47%) in ascending order. Whereas, the rest of wheat varieties under study showed the relatively moderate responses of varietal resistance to the tested Pst isolates during 2017/2018 growing season (Table 7). In the second

season; 2018/19, the highest levels of varietal resistance were found in cvs; Sakha94, Sakha95, Gemmeiza12, Giza171, Sids13 and Misr3 (ranging from 80.00 to 85.00%). In contrast the lowest values were found in cvs; Sids12, Sakha69, Sakha61, Sakha93, Gemmeiza9, Gemmeiza10 Gemmeiza11, Misr1 and Misr2 didn't exceeded up to 40% (Table 7).

**Table 6. Virulence frequency (%) of *Puccinia striiformis tritici* (Pst) isolates corresponding to 34 stripe rust resistance genes (*Yr*'s) in collections during 2017/18 and 2018/19 in Egypt.**

No.	Genotypes	Yr gene	Frequency of virulence (%) of isolates virulent to the indicated Yr gene and gene efficacy(%)					
			2017/2018			2018/2019		
			No.of virulent isolates	Virulence frequency(%)	Gene efficacy(%)	No.of virulent isolates	Virulence frequency(%)	Gene efficacy(%)
1	Avocet R'	<i>YrA</i>	90	69.23	30.67	180	90.00	10.0
2	Yr1/6*Avocet S	<i>Yr1</i>	0	0.00	100.0	0	0.0	100.0
3	Yr5/6*Avocet S	<i>Yr5</i>	0	0.00	100.0	0	0.0	100.0
4	Yr6/6*Avocet S	<i>Yr6</i>	100	76.93	23.07	190	95.0	5.00
5	Yr7/6*Avocet S	<i>Yr7</i>	100	76.93	23.07	190	95.0	5.00
6	Yr8/6*Avocet S	<i>Yr8</i>	80	61.53	38.47	160	80.0	20.0
7	Yr9/6*Avocet S	<i>Yr9</i>	75	57.69	42.31	150	75.0	25.0
8	Yr10/6*Avocet S	<i>Yr10</i>	0	0.00	100.0	0	0.00	100.0
9	Yr15/6*Avocet S	<i>Yr15</i>	0	0.00	100.0	0	0.00	100.0
10	Yr17/6*Avocet S	<i>Yr17</i>	0	0.00	100.0	50	25.00	75.0
11	Yr18/6*Avocet S	<i>Yr18</i>	70	53.84	46.16	105	52.5	57.5
12	Yr27/6*Avocet S	<i>Yr27</i>	0	0.00	100.0	70	35.5	64.5
13	Yr32/6*Avocet S	<i>Yr32</i>	0	0.00	100.0	60	30.0	70.0
14	YrSP/6*Avocet S	<i>YrSP</i>	0	0.00	100.0	40	20.0	80.0
15	Chinese 166	<i>Yr1</i>	0	0.00	100.0	0	0.00	100.0
16	Lee	<i>Yr7</i>	100	76.93	23.07	190	95.00	5.00
17	Heine's Kolben	<i>Yr6+1</i>	70	53.84	46.16	140	70.0	30.0
18	Vilmorin 23	<i>Yr3a, 4a</i>	60	46.15	53.85	120	60.0	40.0
19	Moro	<i>Yr10</i>	0	0.00	100.0	0	0.00	100.0
20	StrubesDickopf	<i>YrSD</i>	60	46.15	61.54	110	55.0	45.0
21	Suwon 92/Omar	<i>YrSU</i>	40	30.76	53.85	120	60.0	40.0
22	Clement	<i>Yr9, Yr2</i>	45	34.6	69.24	80	40.0	60.0
23	Hybrid 46	<i>Yr4</i>	45	34.61	65.39	90	45.0	55.0
24	Reichersberg 42	<i>Yr7+?</i>	100	76.93	23.07	190	95.00	5.0
25	Heine's Peko	<i>Yr6+?</i>	100	76.93	23.07	190	95.0	5.00
26	Nord Desprez	<i>YrND</i>	40	30.08	69.24	70	35.0	65.0
27	Compare	<i>Yr8</i>	40	30.08	69.24	70	35.0	65.0
28	Carstens V	<i>Yr32</i>	0	0.00	100.0	30	15.0	85.0
29	Spalding Prolific	<i>YrSP</i>	0	0.00	100.0	30	15.0	85.0
30	Heines VII	<i>Yr2+?</i>	30	23.07	66.93	60	30.0	70.0
31	Federation4/Kavkaz	<i>Yr9</i>	70	53.84	46.16	140	70.0	30.0
32	Anza	<i>YrA, Yr18</i>	60	46.15	53.85	120	60.0	40.0
33	Kalyansona	<i>Yr2</i>	35	26.92	74.08	80	40.0	60.0
34	Triticum spelta album	<i>Yr5</i>	0	0.00	100	0	0.00	100.0
Total			130			200		

**Table 7. Seedling resistance of 18 local wheat varieties against *Puccinia striiformis tritici* isolates collected from different wheat growing areas in Egypt, during 2017/18 and 2018/19 growing seasons.**

No.	Wheat varieties	Frequency of virulence (%) of Pst isolates virulent to the indicated wheat varieties			
		2017/2018		2018/2019	
		No. of virulent isolates	Varietal resistance(%)	No. of virulent isolates	Varietal resistance(%)
1	Sakha61	60	53.85	120	40.0
2	Sakha69	90	30.77	180	10.0
3	Sakha93	80	38.47	160	20.0
4	Sakha94	10	92.39	30	85.00
5	Sakha95	10	92.39	30	85.00
6	Gemmeiza 9	50	61.54	120	40.0
7	Gemmeiza10	55	56.70	130	35.0
8	Gemmeiza 11	80	38.47	180	10.0
9	Gemmeiza 12	10	92.39	30	85.0
10	Giza 168	30	76.93	50	75.0
11	Giza 171	5	96.16	30	85.0
12	Sids 12	100	23.07	190	5.0
13	Sids 13	80	39.47	40	80.0
14	Sids 14	40	69.24	80	60.0
15	Misr 1	20	74.62	100	50.0
16	Misr 2	10	92.4	130	35.00
17	Misr 3	5	96.16	30	85.0
18	Shandaweel 1	10	92.4	80	60.0
Total		No. of isolates	130	200	

Total of the obtained Pst isolates during each growing season of the study.

**Diversity of pathogen populations under study:**

Three popular indexes of diversity, i.e. Shannon ( $H_{SH}$ ), Gleason ( $H_G$ ) and Simpson ( $H_S$ ), were mainly used to measure the phenotypic variation and the diversity of *Pst* races within populations and between different *Pst* populations, in the tested governorates (populations – regions) under study. These indexes were calculated for each of governorates the six i.e. Kafersheikh, Gharbia, Damietta, Dakahlia, Beheira and Sharqia populations, during the two growing seasons, i.e. 2017/18 and 2018/19 (Table 8 and Fig. 2).

The obtained values of the relative Shannon index as ranged from 0.474 for Sharqia population to 0.554 for Kafersheikh population in the first season, but it was ranged from only 0.392 for Sharqia population to 0.507 for each of Kafersheikh population and Beheira population in the second season. While, Gleason index estimates found to be relatively, wherein it was low less than 0.480 as it was varied between 0.190 and 0.428, in the first season. Also, the estimated value of this index were very small compared with the other two diversity indexes used the smallest of Gleason index between 0.129 and 0.437 in the second season. Moreover, Simpson index values for diversity of races were found in all the tested populations, during the two years of the greatest rather than the other two indices values of the study. Whereas, varied between 0.501 and 0.853 in 2017/18. Also, between 0.528 and 0.820 in 2018/19 (Table, 8).

Comparisons of the different values three indexes of diversity used in this study, for the tested *Pst* populations in various governorates (regions) and during the two years, reflects both variation and/or differences of *Pst* populations in the country. The highest of estimates of Shannon index were observed in Kafersheikh population, followed by Gharbia, Beheira, Damietta, Dakahlia and Sharqia populations. Where, these phenotypic diversity estimates were 0.554, 0.524, 0.509, 0.531, 0.500 and 0.474, in the first season, for the above governorates respectively. Also, *Pst* populations for the previous governorates showed great values of this popular Shannon index where, they were; 0.507, 0.474, 0.474, 0.434, 0.507 and 0.372 in the second season, respectively (Table 8 and Fig. 2).

The Gleason index values of Damietta and Dakahlia populations were relatively low, in comparison with the other populations under study, as it was 0.328 and 0.348 in the first season, but it was 0.333 and 0.337 in the second season for these two populations, respectively. While, the other four pathogen populations of the study i.e. Kafersheikh, Gharbia, Beheira and Sharqia governorates, showed the lowest values of Gleason index, i.e. 0.190, 0.280, 0.217 and 0.230 in 2017/18, while they showed 0.129, 0.171, 0.219 and 0.250 in 2018/19, respectively (Table 8 and Fig. 2).

The highest values of the third popular index of diversity used Simpson index showed, in general the highest values were recorded in the tested *Pst* populations of the six Egyptian governorates; more than 0.500 in both seasons of the study.

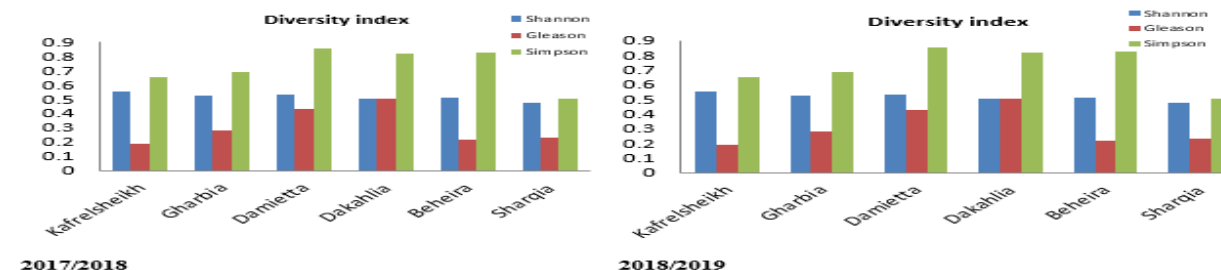
**Table 8. Phenotypic diversity of wheat stripe rust races in collections from six Egyptian governorates, during 2017/18 and 2018/19 growing seasons.**

Governorate	Growing season / Diversity index values					
	2017/2018			2018/2019		
	Shannon	Gleason	Simpson	Shannon	Gleason	Simpson
Kafersheikh	0.554	0.190	0.651	0.507	0.129	0.627
Gharbia	0.524	0.280	0.687	0.474	0.171	0.810
Damietta	0.531	0.328	0.853	0.474	0.333	0.663
Dakahlia	0.500	0.341	0.821	0.434	0.337	0.716
Beheira	0.509	0.217	0.828	0.507	0.219	0.820
Sharqia	0.474	0.230	0.501	0.392	0.250	0.528

In the first season (2017/18), the three *Pst* populations; Damietta, Beheira and Dakahlia showed the highest Simpson index values (up to 0.800); they were 0.853, 0.828 and 0.821, for the previous *Pst* population, respectively. While, the other *Pst* populations under study, Gharbia, Kafersheikh and Sharqia showed also the relatively high values of this index i.e. 0.687, 0.651

and 0.501, respectively. Meanwhile in the second season 2018/19, Simpson index values also, varied between 0.528 in Sharqia population and 0.820 in Bahyria population.

However, the other from populations under study i.e. Kafersheikh, Damietta, Dakahlia and Sharqia revealed in between values of this index i.e. 0.627, 0.663, 0.716 and 0.528, respectively (Table 8 and Fig. 2).



**Fig.2. The Shannon, Gleason and Simpson indexes of phenotypic diversity for 6 populations of *Puccinia striiformis* in Egypt during 2016/17 and 2017/18 growing seasons.**

Relationships between the four components of diversity i.e. size of sample collected from each governorate, number of isolates gained or obtained from

these samples, number of the identified races and standard deviation of race frequency, and each of the three diversity indexes were carried out over the two growing seasons of



the study, and the obtained data presented in Table (9). Significant correlation coefficient were obtained values between each of Shannon and Gleason indexes and the sample size collected from each governorate, the obtained number of isolates, number of the identified *Pst* races and standard deviation of race frequency (Table 9). These results indicated that each of these two popular indexes was sensitive to a corresponding sample size ( $r = 0.540$  &  $0.676$ ), number of isolates ( $r = 0.594$  &  $0.525$ ), number of races ( $r = 0.777$  &  $0.449$ ), and standard deviation of race

frequency ( $r = 0.673$  &  $0.609$ ), respectively. While, very low non correlation coefficient values were present between the above mentioned four components of diversity and the other diversity index under study, i.e. Simpson indexes. So, It can be concluded that the Shannon and Gleason indexes of diversity were the most important indexes to accurately measure the phenotypic diversity of *Puccinia striiformis* races within its populations under Egyptian conditions.

**Table 9. Correlation between independent components of diversity and three diversity indexes of six populations of *Puccinia striiformis* in Egypt, during 2017/18 and 2018/19 growing seasons.**

Diversity indexes	Components of diversity			
	Sample size	No. of isolates	Race number	Standard deviation of race frequency
Shannon ( $H_{SH}$ )	0.540**	0.594**	0.777**	0.673**
Gleason ( $H_G$ )	0.676**	0.525**	0.449**	0.609**
Simpson ( $H_S$ )	0.001 <sup>NS</sup>	0.001 <sup>NS</sup>	0.061 <sup>NS</sup>	0.053 <sup>NS</sup>

**DISCUSSION**

Stripe rust of wheat (*Triticum aestivum*L.), caused by *Puccinia striiformis* f. Sp *tritici* is an economically important disease of wheat in Egypt, as well as many wheat growing areas, worldwide. As, it has been considered the most destructive foliar diseases in the country. The disease could be severely attacked most of the currently used wheat varieties, and caused considerable losses in grain yield, due to their susceptibility to such disease, especially in years when the environmental conditions were favourable for rust incidence and development (Mundt *et al.*, 1995 & Ashamawy and Regab 2016). The observed loss may be reached to approximately 37 % in grain yield of the highly susceptible wheat cultivars in Egypt (Ashmawy and Rageb, 2016). The amount of this loss depends, to a large extent, on the level or degree of aggressiveness of the causal pathogen races, and environmental conditions favorable for disease incidence and development. In this case, the loss in grain yield may reach its maximum and higher levels, specially when the target wheat plants were highly susceptible (Park *et al.*, 1988 and Hong and Singh 1996).

Several yellow rust epidemics have been recorded under Egyptian conditions since 1967. In this season, severe yellow rust infection was occurred on leaves and heads of wheat plants, and destroyed a very large area growing with cultivar; Giza 144, in El-Manzalla district, that located in North Delta of Egypt (Abd el Hak *et al.*, 1972). Also, four epidemics have been reported during the last five decades, i.e. 1983, 1986, 1995 and 1997, and more recently in 2018/19 growing season. However, these epidemics were, in fact the main cause for eliminating and discarding the widely grown and high yielding wheat cultivars from the commercial productions in the field such as; Gemmeiza 1-3 and 5, Giza163, Sakha69, long spikes; Sids cultivars and recently Sakha 93. (Abu El-Naga *et al.*, 1999 and Ashmawy *et al.*,2012)

Field observations during the annual survey of wheat rusts in Egypt, during the two years of the study, revealed, in general, that little or few stripe rust infection was observed in the first growing season; 2017/18, compared to that occurred in the second season; 2018/19. However, relatively late stripe rust infection was recorded in this season of the study, as the disease appeared late in March, 2018 on the plants of bread wheat cv. Sids12,

growing in Kafrelsheikh governorate. Also, the environmental conditions were found to be unfavourable for the disease development and increase during this season. Which, in turn, delayed and slow down the rate of disease, increase thus reduced the amount of the disease, and minimized its severity (%) in host plants of the most wheat growing cultivars all over the country. In contrast, severe stripe rust epidemic have been early recorded in the second season; 2018/19. Due to the early appearance of stripe rust infection in this season, as it was firstly detected in 30 January on the plants of the two wheat cultivars; Sids 12 and Gemmeiza1, that widely grown in the Northern governorates, including Kafrelsheikh. Beside to the early detection of both initial infection and time of outbreak, the high amount of rainfall, combined with the lower degrees of average minimum and maximum temperatures, recorded during this season, all of these reasons significantly contributed to the establishment, spread, and subsequent epidemic outbreak of stripe rust.

During this season, it was also noticed that almost all wheat cultivars widely grown in the country were susceptible to the sudden emergency and of the new and more aggressive *Pst* pathotypes that was initially identified and firstly appeared in pathogen population, that contributed to the occurrence of this epidemic. Stripe rust inoculum had been rapidly disseminated and widely distributed to the other parts of wheat growing areas of the country, nationwide.

Although, the dynamic nature of wheat rust pathogens, host-genetic resistance or growing resistant cultivars, is still the most effective method to control stripe rust of wheat. The failure of genetic resistance in some wheat cultivars after its new release and wide cultivation in the commercial fields was mainly due to the high evolutionary potential of stripe rust populations (McVey *et al.*, 2004).

However, the dynamic nature of such pathogen led to continuous producing and sudden emergency of new virulent and more aggressive races, that can rapidly increase in their frequency and capable to overcome or breakdown resistance genes newly deployed in the recommended wheat cultivars. Therefore, genetic resistance of these newly released wheat cultivars subjected to a rapid loss in its efficacy, and these cultivars become susceptible in a short period of time (few years) or

short duration. Therefore, it is essential to an early detection of the first existence of new races in its pathogen population, before it becomes more prevalence, more frequency and able to cause a significant loss. As, it was reported in the previous studies that there is a relatively long period of time (about 3 to 5 years) from the first detection of a new race in its population to cause a significant crop loss (Chen *et al.*, 2009).

Successful breeding program for rust resistance, especially wheat stripe rust, requires a full understanding of the genetic structure of the causal pathogen populations, and the probable changes occurring in these populations, *i.e.* new races present, their frequencies of virulence and their occurrence and diversity within different geographical areas in the country .

In the course of this study, virulence survey was carried out by collecting stripe rust samples from different locations of the country *i.e.* Kaferelsheikh, Gharbia, Damietta, Dakahlia, Beheria and Sharqia. The results obtained were established on the comparisons of both visual symptoms as infection types (IT's) of the uredial stage of the casual pathogen with those reported by Wiese (1977) and Agrios (1979). Isolation and identification of the prevailing races of the disease are an essential step to satisfy this work (Chen *et al.*, 2002)

In the present study, stripe rust (Pst) race; 64E0 was the most common with consistently high frequency of 26.92%, in its population, during 2017/18 growing season. Meanwhile, race 450E254 was the most dominant Pst race, as it represented by 22.5% frequency in 2018/19 growing season. Similar results were previously obtained by Nazim *et al.*, (2010), as they previously reported that the two stripe rust races; 0E0 and 102E22, were the most frequent in Egypt during the two growing seasons of their study; 2005/06 and 2006/07. Also, Shahin *et al.*, (2015) found, under the Egyptian conditions that stripe rust races; 0E0 and 6E16 were the most frequent (each with 12.54% frequency) during 2013/14. While, the three races; 0E0, 4E8 and 198E154 were the most common races (with 12.16, 9.15 and 8.45% frequencies, respectively) during 2014/15 growing season

Results obtained in the current study, confirmed by the previous studies of Nazim *et al.*, (2010), and Shahin *et al.*, (2015), supported the conclusion that wheat stripe rust population in Egypt, has been dominated by a few pathogen races (only one or two races), each year during their studies. Also, these common races did not reappear year after year in pathogen population, under the Egyptian conditions.

Dominance by a few races may be expected in pathogen populations, those maintain the relatively small population sizes, and are not subject to loss of some races in episodes of population crashes (genotype drift), between the successive growing seasons of the host crop. If the common Pst races could not occur in its pathogen populations each year, it is likely to suggest that those races did not survive the summer in Egypt and the source of airborne urediniospores (primary inoculum) is consistent from year to year in an external source and/or an adjacent country that provides a rich diversity of races each year Kolmer *et al.*,(2005)

If Pst fungal pathogen propagules not oversummers in Egypt, it could be possible to maintain long term and sustainable effectiveness of race – specific resistance in the Egyptian wheat cvs; but care is taken to avoid use of the same resistance genes, that are widely used in the source region of the primary inoculum. (Abdel-Hak *et al.*, 1974; Nazim *et al.*, 1976; McVey *et al.*, 2004 and Negm *et al.*, 2013).

The obtained data in the current study showed, on the other hand, that Pst populations of Kaferelsheika governorate were the largest population sizes, as they have the highest number of the identified stripe rust races. Wherein, they represented 21.53% and 20.5% frequencies of the total races in the annual Egyptian populations during, 2017/18 and 2018/19 growing seasons, Followed by Gharbia and Sharqia populations, where they also showed the relatively high frequencies of the total races in population. While, the three governorates; Dakahlia, Beheira and Damietta showed the lowest number of the identified Pst races in their pathogen population.

On the other hand, the three Pst races *i.e.* 64E0, 66E0 and 70E26 are the most widespread races in all locations, nationwide, as each was found in six governorates in 2017/18. While, the other three Pst races; 192E192, 206E254 and 450E254 are the most geographically distributed races, which was recorded in six governorates under study. Similar results were previously obtained by Shahin *et al.*, (2015), Nazim *et al.*, (2010), Ashmawy (2010) and Sajid *et al.*, (2014).

The obvious Shifts or changes in race structure of Pst annual populations in Egypt, could be confirmed by the relationship between them and alterations in virulence frequencies against number of *Yr's* genes. Analyzing and testing virulence of Pst isolates to stripe rust resistance genes (*Yr's*), combined with the genetic structure of Pst populations, have provided valuable data and/or an important information for wheat breeding program. In this study, no virulences were occurred corresponding to the four *Yr's*; *Yr1*, *Yr5*, *Yr10* and *Yr15* against the tested isolates. These *Yr's* considered to be of highly effective (completely resistant) against all the obtained *Pst* isolates, during the two years of the study. According to the highest virulence frequencies, to *Yr's*; *Yr17*, *Yr27*, *Yr3*, *Yr6*, and *Yr7*, (more than 80%), these stripe rust resistance genes considered to be ineffective genes under Egyptian conditions during the two years of the study. The previous study of Shahin (2017), evaluated 47 differential sets of monogenic lines with known yellow rust resistance genes under Egyptian field conditions. This study indicated that, out of the tested genes only 10 *Yr* genes were resistant to stripe rust pathogen at adult plant stage, as they showed rust response ranged from 0 – 5MR. Another study of Chen *et al.*, (2009) reported that the yellow rust resistance genes; *Yr5*, *Yr10*, *Yr15* and *Yr24/26* confer resistance to Pst race; CYR32 under Chinese conditions.

In the current study, different disease response to the collected stripe rust isolates were observed on the tested wheat cultivars, suggested that these wheat genotypes had diverse genetic backgrounds and having various *Yr* resistance genes. The highest levels of varietal resistance to the tested Pst isolates were displayed by the wheat cultivars *i.e.* Sakha 94, Sakha 95, Gemmeiza12, Misr2, Shandaweel 1, Giza 171

and Misr 3, in 2017/2018 growing season. While, out of the previous cultivars, only three cvs., Sakha 95, Gemmeiza 12, and Misr 3, showed the highest levels of varietal resistance in the second season; 2018/19. On the other hand, virulence to wheat cvs., Sids 12, Sakha 69, Gemmeiza 9, Gemmeiza 10, Gemmeiza 11, Sakha 93, Misr 1 and Misr 2, the lowest was found to be occurred at greater than 90% frequency in the two years under study. Similar results were previously reported by the findings of Shahin *et al.*, (2018).

Diversity indexes are the main quantitative measurements that accurately reflect how many different types (such as plant pathogen races) found in a community or in a population, and simultaneously take into account how these races are distributed among those types (Andriveau and de Vallavieille-Pope, 1995). The measurement of diversity between and/or within any pathogen population is one of its important characteristics. As previously reported by Groth and Roelfs (1987) an optimal diversity index should satisfy several conditions: A) pathogen population is more diverse (showed higher index values), if it consists of a larger number of races, also, when it is characterized by an even distribution of races. In contrast, the low diversity alternative being the case where a small or a few number of races were dominant in the population, and all other races being rare, with low frequencies in their populations.

In this part of the study, three popular and widely used diversity indexes, i.e. Shannon, Simpson and Gleason were mainly used to accurately estimate and measure phenotypic diversity of *Puccinia striiformis* populations in six different locations in Egypt. The relative Shannon index showed low values that differed from the other two indexes, i.e. Simpson and Gleason. This is mainly due to the lowest number of isolates in most populations and low values of frequency (%) of most races in their populations. While, both Gleason and Simpson indexes showed, in general, high values, compared with the other diversity indexes under study. This means that high diversity was found for the studied populations in most of the tested locations, during the two growing seasons. The high diversity in different stripe rust populations under study, could be explained by the wide cultivation of the commercial Egyptian wheat cultivars for a long period of time (many years), having high levels of stripe rust resistance. This makes high selection pressure on *Pst* races, imposed by the major *Yr* resistance gene(s), newly deployed in the Egyptian wheat cultivars. This issue was previously supported by the findings of Singh *et al.*, (2004). Similar results were previously obtained by Hovmoller *et al.*, (2011), Bux *et al.*, (2012), Sajid *et al.* (2014) and Prashar *et al.*, (2015).

Further studies are required or needed in the future for surveying and monitoring the dynamics and virulence diversity of stripe rust races in its populations between years and wheat growing areas in different governorates in Egypt, using monogenic lines for stripe rust resistance genes (*Yr*'s), as differentials, in combination with molecular marker characterization of *Pst* races, to provide an essential basis that are very needed or of great importance for an anticipatory breeding program aimed to produce new wheat cultivars with a sustainable resistance to stripe rust.

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#### REFERENCES

- Abd El-Hak, T.M.; Stewart, D.M. and Kamel, A.H. (1972). The current rust situation in the near East countries. Regional Wheat Workshop. Beirut Lebanon, 1-29.11-15:81-90.
- Abdel-Hak, T.; El-Shehedi, A.A. and Nazim, M. (1974). The source of inoculum of wheat leaf rust in relation to wind direction in Egypt. *Egypt J. Phytopathol.*, 6:17-25.
- Abu El-Naga, S.A.; Khalifa, M.M.; Sherif, S.; Youssef, W.A.; El-Daoudi, Y.H. and Ikhlas Shafik, E.H. (2001). Virulence of wheat stripe rust pathotypes identified in Egypt during 1999/2000 and sources of resistance. First Regional Yellow Rust Conference for Central & West Asia and North Africa 8-14 May, SPH, Karj, Iran.
- Abu-El-Naga, S.A.; Khalifa, M.M.; Bassiouni, A.A.; Youssef, W.A.; Shehab El-Din, T.M. and Abd-Latif, H.A. (1999). Revised evaluation for Egyptian wheat germplasm against physiologic pathotypes of stripe rust. *J. Agric. Sci. Mansoura. Univ.*, 24 (2) : 477 – 488
- Agrios, G. N. (1979). *Plant Pathology*, Academic Press, New York, pp.703.
- Andriveau, D. and deVallavieille- Pope, C. (1995). Race diversity and complexity in selected populations of fungal biotrophic pathogens of cereals. *Phytopathology*, 85:897-905.
- Ashmawy, M. A. (2010). Advanced studies on stripe rust of wheat in Egypt. Msc. Thesis, a Botany Department Faculty of Agriculture, Minufiya University Shebin ELkom pp.
- Ashmawy, M. A. and Ragab, Kh. E. (2016). Grain yield of some wheat genotypes to stripe rust in Egypt. *Menoufia J. Plant Prot.*, Vol. (1) : 9- 18.
- Ashmawy, M. A. ; Abu Aly, A. A. ; Yousef, W. A. and Shahin, A. A. (2012). Physiologic races of wheat yellow rust (*Puccinia striiformis* f. sp. *tritici*) in Egypt during 1999- 2011., *Menoufia J. Agric. Res.* 37(2) :297- 305.
- Bux, H. ; Rasheed, A. Mangrio, S.M. ; Abro, S.A. Shah, S.J. ; Ashraf, M. and Chen, X. (2012). Comparative virulence and molecular diversity of stripe rust *Puccinia striiformis* f. sp. *tritici* collections from Pakistan and United states. *Int. J. Agric., Biology* Vol. 14 No. 6 : 851- 860.
- Chen, W. Q.; Wu, L.R.; Liu T.G.; Xu S.C.; Jin S.L.; Peng Y.L. and Wang, B.T. (2009). Race dynamics, diversity, and virulence evolution in *Puccinia striiformis* f. sp. *tritici*, the causal agent to wheat stripe rust in China from 2003 to 2007. *Plant Dis.*, 93: 1093-1101. doi: 10.1094/PDIS-93-11-1093
- Chen, X. M.; Moore, M.; Milus, E. A.; Long, D. L.; Line, R. F.; Marshall, D. and Jackson, L. (2002). Wheat stripe rust epidemics and races of *P. striiformis* f. sp. *tritici* in the United States in 2000. *Plant Disease* 86:1, 39-46.
- El-Daoudi, Y. H.; Ikhlas Shafik, E. H.; Ghanem, S.; Abu El-Naga, S. A.; Sherif, S. O.; Khalifa, M. M.; Mitkees, R. A. and Bassiouni, A.A. (1996). Stripe rust occurrence in Egypt and assessment of grain yield loss in 1995. Proc. Du Symposium Regional Sur les Maladies des Cereales et des Legumineuses Alimentaries 11-14 November, 1996, Rabat, Maroc.

- Groth, J.V. and Roelfs, A.P. (1987). The concept and measurement of phenotypic diversity in *Puccinia graminis* on wheat. *Phytopathology*, 77:1395-1399.
- Hong, M.A. and Singh, R.P. (1996). Contribution of adult plant resistance gene yr18 in protecting wheat from yellow rust. *Plant Disease* 1: 66-69.
- Hovmoller, M. S.; Sorencen, Ch.; Nalter, S. and Justesen, A. F. (2011). Diversity of *Puccinia Striiformis* on cereals and grasses. *Annu. Rev. Phyto. Patho.* 49 : 197- 217.
- Jin, Y.; Szabo, L.J. and Carson, M. (2010). Century-old mystery of *Puccinia striiformis* life history solved with the identification of *Berberis* as an alternate host. *Phytopathology*,100:432-435
- Johnson, R.; Stubbs, R. W.; Fuch, E. and Chamberlain, N. H. (1972). Nomenclature for physiologic races of *P. striiformis* infected wheat. *Tran. Br. Mycol. Soc.*, 58: 475-480. (C.F.) Knott (1989).
- Kolmer, J.A. (2005). Tracking wheat rust on a continental scale. Oxford, U.K.; El Sevier. *Current Opinion in Plant Biology*, 8:441-449.
- McNeal, F. H.;Konzak, C.S. ; Smith, E. P.; Tate, W. S. and Russel, T.S. (1971). A uniform system for recording and process in cereal data. *USDA ARS* 34-121.
- McVey, D.V.; Nazim, M.; Leonard, K.J. and Long, D.L. (2004). Patterns of virulence diversity in *Puccinia triticina* on wheat in Egypt and the United States in 1998-2000. *Plant Dis.*, 88:271-279.
- Morgounov, A.; Gummadov, N.; Belen, S.; Kaya, Y.; Kesfr, M. and Mursalova, H. (2014) Association of digital photo parameters and NDUI with winter wheat grain yield in variable environments, *Turk. J. Agric.* 38: 624-632.
- Mundt, C.C.;Brophy, L.S. and Schmitt, M.S. (1995).Disease severity and yield of pure- line wheat cultivars and mixture in the presence of eyespot, yellow rust, and their combination. *Plant Pathology* 44: 173 – 182
- Nazim, M.; El-Shehedi, A.A.; El-Basyouni, S.Z. and Abdel-Hak, T.M. (1976). The relative effectiveness of wheat leaf rust resistance genes in seedling and adult stages. 2<sup>nd</sup> Cong. Egypt. *Phytopathology Soc.*, Pp. 627-641.
- Nazim, M; Awad, M.A.; Khalifa, S.Z.; Abu El-Naga, S.E., and Ashmawy, M. A. (2010). Fréquence of virulence and virulence formula of wheat stripe rust races identified in Egypt. *Menoufia J. Agric. Res.* Vol. 35 No 2: 439- 452.
- Negm, S.S.; Boulot, O.A. and Hermas, G.A. (2013). Virulence dynamics and diversity in wheat leaf rust (*Puccinia triticina*) populations in Egypt during 2009/2010 and 2010/2011 growing seasons. *Egypt. J. Appl. Sci.*, 28:183-212.
- Park, R.F.; Rees, R.G. and Platz, G.J. (1988). Some effects of stripe rust infection in wheat with adult plant resistance. *Australian Journal of Agricultural Research* 39,(4): 555-562.
- Prashar, M.;Bhardwaj, S.C. ; Jain, S.K. ; and Gangwar. (2015). Virulence diversity in *Puccinia striiformis* f. sp. *tritici* causing yellow rust on wheat (*Triticum aestivum*) in India., *Indian Phytopathol* 68 (2) : 129-133.
- Sajid, A.; Leconte, M.; Rahman, H. ; Saqib, M.S.; and Gladieux, P. (2014). A high virulence and pathotype diversity of *Puccinia Striiformis* f. sp. *tritici* its center diversity, the Himalayan region of Pakistan *Eur. J. Plant Pathol.* 140: 275- 290.
- Shahin A.A. 2017. Effective genes for resistance to wheat yellow rust and virulence of *Puccinia striiformis* f. sp. *tritici* in Egypt. *Egypt. Acad. J. Biolog. Sci.*, 8(2): 1–10.
- Shahin A.A.; Abu Aly, A.A. and Shahin, S.I. (2015). Virulence and diversity of wheat stripe rust pathogen in Egypt. *Journal of American Science*, 11(6):47-52.
- Shahin A.A.; and Hend A. Omar and El-Sayed, A.B. (2018). Characterization of *Yr18/Lr34* partial resistance gene to yellow rust in some Egyptian wheat cultivars, *Egy. J. Plant Pro. Res.* 6(3):1-9.
- Singh, R.P.; Huerta-Espino, J.; Pfeiffer, W. and Figueroa Lopez, P. (2004). Occurrence and impact of a new leaf rust race on durum wheat in North western Mexico from 2001 to 2003. *Plant Dis.*, 88:703-708.
- Stakman, E.C.; Stewart D.M. and Loegering W.Q.(1962) Identification of physiologic races of *Puccinia graminis tritici* ARS, USDA, Agr. Res. Serv.Bull.E-617. 53 pp.
- Wiese, M.V. (1977). Compendium of Wheat Disease, Incorporation with Dept. Bot. and Plant Pathology, Michigan State Univ. of Nebraska, Lincoln. pp 106.

## ديناميكية القدرة المرضية والتنوع في عشائر الفطر *Puccinia striiformis tritici* في مصر خلال موسمي 2017/18 و 2018/19

مدوح عبدالمنعم عشموى ، عاطف عبدالفتاح شاهين ، سمر محمد عادل إسماعيل و هند عبدالنبي محمد  
مركز البحوث الزراعية ، معهد بحوث امراض النباتات ، قسم بحوث امراض القمح ، مصر.

يعتبر مرض الصدا المخطط المتسبب عن الفطر بكسينيا سترايفورمس من الامراض ذات الأهمية الاقتصادية والتي تصيب القمح في مصر . لذلك فقد أجرى هذا البحث بهدف دراسة ديناميكية القدرة المرضية والتنوع في عشائر الفطر المسبب للمرض في مصر خلال موسمي الزراعة 18 / 2017 و 19 / 2018 . ولتحديد التركيب الجيني لعشائر الفطر المسبب تم تحليل ودراسة 330 عزلة ممرضة من الفطر المسبب تم الحصول عليها من 250 عينة نباتية مصابة بالمرض أخذت من ستة محافظات وهي (كفر الشيخ ، دمياط ، الشرقية، البحيرة ، الدقهلية ، والغربية) وفي خلال تلك الدراسة تم تعريف 9 سلالات فطرية في الموسم الأول وكذلك 10 سلالات فطرية في الموسم الثاني حيث كانت السلالة 64E0 أكثر تلك السلالات شيوعا في عشيرة الفطر المسبب يليها في ذلك السلالتين 66E0 (بتكرار 23.07%) ثم السلالة 70E26 (بتكرار قدره 15.38%) وذلك خلال موسم الزراعة الأول بينما كانت السلالة الفطرية 450E254 أكثر تلك السلالات تكرارا داخل عشيرة الفطر وذلك بتكرار قدره 22.5% يليها في ذلك اربع سلالات هم 206E174 ، 192E192 ، 136E54 و 70E182 بتكرارا قدرها 10.0 ، 10.0 ، 15.0 ، 17.5 % على التوالي خلال موسم الزراعة 2018 / 2019 . وقد أظهرت النتائج المتحصل عليها بتلك الدراسة أن السلالات 64E0، 66E0، 70E22 كانت الأكثر انتشارا بين المحافظات تحت الدراسة وذلك خلال موسم الدراسة الأول بينما كانت السلالات 136E54 ، 192E192 ، 206E174 أكثر تلك السلالات انتشارا في جميع المحافظات تحت الدراسة في الموسم التالي وبمقارنة التشابه والاختلاف بين عشائر الفطر المسبب للمرض في المناطق المختلفة أتضح أن هناك درجة عالية من التشابه بين تلك العشائر في المحافظات تحت الدراسة خلال موسمي الزراعة . ومن ناحية أخرى فقد تم قياس التنوع والتباين بين عشائر الفطر الممرض وذلك باستخدام ثلاثة مؤشرات أساسية للتباين وهم: Shannon و Geleason حيث أوضحت النتائج المتحصل عليها أن مؤشر Shannon كان أكثر تلك المؤشرات ثباتاً ودلالة على قياس التنوع في عشائر الفطر المختلفة حيث اتضح أن هذا المؤشر كان أكثرهم تأثراً بحجم العينة وعدد العزلات وعدد السلالات . ويعتبر قياس التنوع وديناميكية القدرة المرضية في عشائر الفطر(المخطط) المسبب لمرض الصدا الأصفر أحد المتطلبات الأولية الهامة لإجراء المقارنه لكونها تعد احد القواعد الأساسية التي تعتمد عليها المربي لإجراء البرنامج المستقبلي للتربية لمقاومة ذلك المرض في برامج التربية في مصر .